# American POTATO JOURNAL

*Volume 29* *February, 1952* *Number 2*

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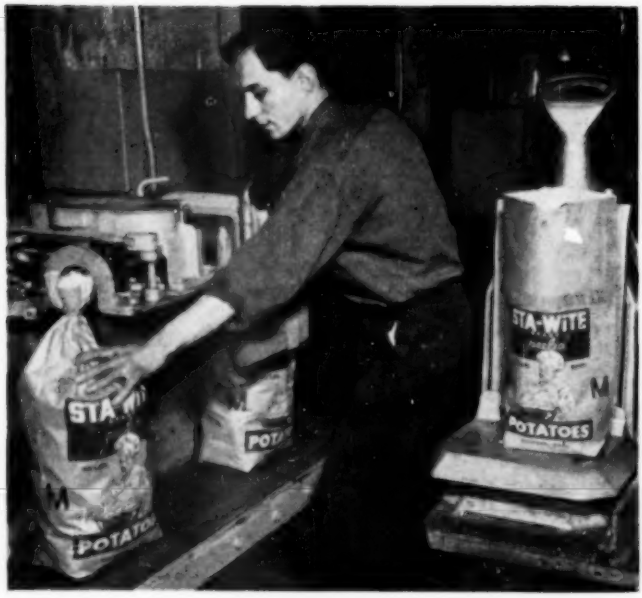


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# American Potato Journal

PUBLISHED BY
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NEW BRUNSWICK, N. J.



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Entered as second class matter at New Brunswick, N. J., March 14, 1942 under Act of March 3, 1879. Accepted for mailing at special rate of postage provided for in section 412, Act of February 28, 1925, authorized on March 14, 1928.


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## VARIATIONS IN SPECIFIC GRAVITY OF POTATOES[1]


P. H. HEINZE[2] AND C. C. CRAFT[3]
*Bureau of Plant Industry, Soils and Agricultural Engineering, Beltsville, Md.*

AND

BEATRICE M. MOUNTJOY AND MARY E. KIRKPATRICK[4,5]
*Bureau of Human Nutrition and Home Economics, United States Department of Agriculture, Beltsville, Md.*



(Accepted for publication September 24, 1951)


Many factors such as rainfall, temperature, sunlight, soil type, fertilizers, irrigation, and more recently the use of fungicides, insecticides and vine killers have been listed as influencing the specific gravity of potatoes (2, 5, 6). That different varieties and kinds of potatoes vary in specific

[1] This work is a part of a cooperative project between the Bureau of Plant Industry, Soils, and Agricultural Engineering and the Bureau of Human Nutrition and Home Economics on Predetermining and Evaluating the Cooking Quality of Potatoes and was made under the Research and Marketing Act of 1946 #147.

[2] Physiologist.

[3] Assistant Physiologist.

[4] Food Specialists.


[5] The authors acknowledge the assistance of E. J. Koch, Bureau of Plant Industry, Soils and Agricultural Engineering in the statistical analysis of the data.

gravity and hence in food value has been recognized for over 100 years (7).

The relation between specific gravity and certain cooking quality factors, such as mealiness, has been noted by a number of investigators. The existence of considerable variation in specific gravity of the individual tubers in small lots of potatoes has aroused interest in the possibility of commercial separation of potatoes into lots of different specific gravities. Potato breeders (1) consider specific gravity a definite genetic characteristic although considerable differences are found among tubers of the same variety in the same field. The purpose of this paper is to report the variations in specific gravity that occurred among the tubers in several varieties from different growing areas over a period of several years and to note the influence of some storage conditions on the specific gravity of the potatoes.

## Materials and Methods

Five varieties each from three locations and one variety (Triumph) from two locations were used. The lots were obtained from commercial production areas in ten different states but were not necessarily from the same farms or fields in the different years. All were graded U. S. No. 1 Size A unwashed late crop potatoes. Specific gravity was determined by immersion of the individual tubers in salt solutions of different concentrations as described by Clark *et al.* (3). Fifty-tuber samples were used from the freshly harvested lots and various other samples were taken after 2 to 4 months' storage (midseason storage), at 40°F. and 55°F. Other samples were taken after 5 to 6½ months' storage (late season storage), at 40°F. Temperatures in the storage rooms were thermostatically controlled, but the humidities were regulated largely by manual devices. Attempts were made to maintain relative humidities of 85 to 90 per cent, but there was some variation.

## Discussion of Results

*Standard Deviations of Specific Gravity.*

The total number of tubers tested, the average specific gravity for each lot computed from the values for all the individual tubers and 2 times the standard deviations are given in table 1 for each variety at its specific location for each year. The average specific gravity differs from year to year for the lots obtained from the same general location. The deviations indicate that these same lots were much more variable in some years than in others. The weighted average deviations for location show that potatoes tend to be more variable in specific gravity in some locations. For example, in the data for the Katahdin variety, high deviations are shown in the sample from Colorado. The Irish Cobbler and Chippewa varieties were more uniform in specific gravity than the other varieties. This indicates that commercial separation on the basis of specific gravity

is more feasible with some lots of potatoes than with others. It must be emphasized that all varieties were not grown in the same locations therefore the differences noted for varieties may be influenced considerably by location.

Grieg (4) has shown in a recent experimental study that consumers are interested in buying potatoes separated into several different specific gravity lots and marketed for different cooking purposes. The variability between varieties and between locations of each variety shown in the data reported here reemphasizes the need for such a separation in order to make the best possible use of potatoes in cooking.

*Effect of Storage upon Specific Gravity*

The changes in specific gravity during storage are given in table 2. In 1948 the averages for all varieties showed no appreciable change at 55°F. between harvest and the midseason storage period. However, tubers stored at 40°F. showed an increase in specific gravity by midseason and further increase in the late season storage period. In 1949 the averages for all varieties showed an increase in specific gravity for those lots stored at 55°F. but very little change for those stored at 40°. When these values are compared with the average relative humidity values calculated from hygrothermograph-records it is easily seen that storage at the lower relative humidities (83 to 84 per cent) caused an increase in specific gravity in some varieties. Very little change occurred when the relative humidity was maintained at approximately 90 per cent. Terman, Goven and Cunningham (8) reported an increase in specific gravity in each of four varieties during storage at three different temperatures but they did not report the humidity conditions at which the potatoes were stored. A comparison of specific gravity values for individual varieties shows that the greatest increase in specific gravity occurred in the Katahdin and Chippewa varieties whereas Irish Cobbler showed a decrease in all lots except those stored at 55° in 1949. Triumph and Green Mountain follow Irish Cobbler in rank as showing less change in specific gravity during storage than other varieties. The Russet Burbank lots were intermediate in most instances.

The various lots of the 1949 crop were weighed at harvest and at sampling periods and the per cent loss in weight was calculated (Table 3). Shrinkage was found to be closely related to changes in specific gravity. The Irish Cobbler variety showed the lowest shrinkage with a maximum of 4.1 per cent. With the exception of a 7.4 per cent shrinkage for the Green Mountain lot at 55°F. the Chippewa lots showed the highest shrinkage for all storage periods. These data agree well with the frequently observed greater turgidity of the Irish Cobbler potatoes as compared with many other varieties after prolonged storage.

TABLE 1.—*Average specific gravity of potatoes and standard deviations of average values for potatoes from different locations and of different varieties*

| Variety | Location | Year | Number of Tubers | Average Specific Gravity | 2 Times Standard Deviation[1] | | |
|---|---|---|---|---|---|---|---|
| | | | | | Individual Lot Value | Weighted Average for Location[2] | Weighted Average for Variety[2] |
| Chippewa | Indiana | 1948 | 456 | 1.067 | .016 | | .013 |
| | | 1949 | 556 | 1.065 | .012 | .014 | |
| | Maine | 1947 | 100 | 1.069 | .009 | | |
| | | 1948 | 481 | 1.083 | .015 | | |
| | | 1949 | 593 | 1.069 | .010 | | |
| | | 1950 | 162 | 1.066 | .009 | .012 | |
| | Michigan | 1947 | 99 | 1.070 | .011 | | |
| | | 1948 | 430 | 1.082 | .017 | | |
| | | 1949 | 546 | 1.076 | .013 | | |
| | | 1950 | 168 | 1.074 | .012 | .013 | |
| Green Mountain | Maine | 1947 | 92 | 1.086 | .014 | | .015 |
| | Heavy Soil | 1948 | 471 | 1.085 | .018 | | |
| | | 1949 | 594 | 1.078 | .012 | | |
| | | 1950 | 179 | 1.081 | .017 | .015 | |
| | Maine | 1947 | 100 | 1.082 | .012 | | |
| | Light Soil | 1948 | 472 | 1.094 | .017 | | |
| | | 1949 | 619 | 1.089 | .013 | | |
| | | 1950 | 103 | 1.083 | .013 | .014 | |
| | New York | 1947 | 99 | 1.099 | .017 | | |
| | Long Island | 1948 | 459 | 1.096 | .014 | | |
| | | 1949 | 645 | 1.081 | .017 | | |
| | | 1950 | 175 | 1.088 | .014 | .016 | |
| Irish Cobbler | Maine | 1947 | 100 | 1.081 | .012 | | .013 |
| | | 1948 | 477 | 1.087 | .014 | | |
| | | 1949 | 611 | 1.087 | .014 | | |
| | | 1950 | 126 | 1.078 | .010 | .013 | |
| | North Dakota | 1948 | 477 | 1.087 | .014 | | |
| | | 1949 | 571 | 1.090 | .012 | | |
| | | 1950 | 120 | 1.084 | .014 | .013 | |
| | Wisconsin | 1947 | 99 | 1.081 | .011 | | |
| | | 1948 | 470 | 1.082 | .013 | | |
| | | 1949 | 585 | 1.077 | .012 | | |
| | | 1950 | 105 | 1.081 | .014 | .012 | |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| Katahdin | | | | | | | .016 |
| | Colorado | 1947 | 100 | 1.081 | .016 | | |
| | | 1948 | 480 | 1.094 | .018 | | |
| | | 1949 | 615 | 1.092 | .018 | | |
| | | 1950 | 99 | 1.090 | .022 | .018 | |
| | Maine | 1947 | 100 | 1.077 | .011 | | |
| | | 1948 | 474 | 1.089 | .016 | | |
| | | 1949 | 600 | 1.078 | .014 | | |
| | | 1950 | 102 | 1.081 | .011 | .014 | |
| | Pennsylvania | 1947 | 100 | 1.067 | .012 | | |
| | | 1948 | 440 | 1.077 | .018 | | |
| | | 1949 | 566 | 1.071 | .016 | | |
| | | 1950 | 92 | 1.070 | .011 | .016 | |
| Russet Burbank | | | | | | | .014 |
| | Idaho | 1947 | 99 | 1.096 | .020 | | |
| | Irrigated | 1948 | 462 | 1.089 | .014 | | |
| | | 1949 | 580 | 1.089 | .012 | | |
| | | 1950 | 110 | 1.091 | .012 | .013 | |
| | Idaho | 1948 | 468 | 1.096 | .015 | | |
| | Non-irrigated | 1949 | 561 | 1.086 | .012 | | |
| | | 1950 | 142 | 1.088 | .012 | .013 | |
| | Washington | 1947 | 102 | 1.102 | .010 | | |
| | | 1948 | 466 | 1.095 | .018 | | |
| | | 1949 | 509 | 1.098 | .012 | | |
| | | 1950 | 108 | 1.088 | .012 | .014 | |
| Triumph | | | | | | | .014 |
| | North Dakota | 1947 | 99 | 1.088 | .014 | | |
| | Heavy Soil | 1948 | 469 | 1.083 | .016 | | |
| | | 1949 | 566 | 1.086 | .013 | | |
| | | 1950 | 121 | 1.077 | .010 | .014 | |
| | North Dakota | 1948 | 472 | 1.094 | .016 | | |
| | Light Soil | 1949 | 548 | 1.081 | .011 | | |
| | | 1950 | 178 | 1.088 | .014 | .013 | |

[1] 2 times standard deviation includes 95.45 per cent of the tubers in each lot.
[2] These averages weighted according to number of tubers in each lot.

TABLE 2.—*Effect of storage temperature and humidity on the specific gravity of six varieties of potatoes*

| Variety | 1948 | | | | 1949 | | | |
|---|---|---|---|---|---|---|---|---|
| | Harvest | Midseason Storage[1] | | Late Season Storage[2] | Harvest | Midseason Storage[1] | | Late Season Storage[2] |
| | | 55°F. | 40°F. | 40°F. | | 55°F. | 40°F. | 40°F. |
| Chippewa | 1.074 | 1.074 | 1.077 | 1.082 | 1.068 | 1.071 | 1.070 | 1.071 |
| Green Mountain | 1.090 | 1.088 | 1.092 | 1.096 | 1.082 | 1.085 | 1.082 | 1.082 |
| Irish Cobbler | 1.086 | 1.083 | 1.083 | 1.085 | 1.086 | 1.086 | 1.084 | 1.083 |
| Katahdin | 1.083 | 1.085 | 1.087 | 1.090 | 1.078 | 1.082 | 1.080 | 1.080 |
| Russet Burbank | 1.090 | 1.091 | 1.093 | 1.097 | 1.090 | 1.092 | 1.090 | 1.092 |
| Triumph | 1.087 | 1.087 | 1.089 | 1.090 | 1.082 | 1.086 | 1.083 | 1.084 |
| Average All Varieties | 1.085 | 1.085 | 1.087 | 1.090 | 1.081 | 1.084 | 1.081 | 1.082 |
| Per cent Relative Humidity | | 90 | 83 | 83 | | 84 | 88 | 88 |

[1] 2 to 4 months
[2] 5 to 6½ months

TABLE 3.—*Loss in weight of potatoes of the 1949 crop from harvest through storage*

| Variety | Midseason Storage[1] | | Late Season Storage[2] |
|---|---|---|---|
| | 55°F. | 40°F. | 40°F. |
| | Per cent | Per cent | Per cent |
| Chippewa | 7.3 | 4.3 | 7.3 |
| Green Mountain | 7.4 | 3.9 | 5.1 |
| Irish Cobbler | 4.1 | 2.8 | 3.2 |
| Katahdin | 6.9 | 3.9 | 6.7 |
| Russet Burbank | 4.2 | 3.0 | 4.0 |
| Triumph | 4.7 | 2.8 | 5.2 |

[1] 2 to 4 months
[2] 5 to 6½ months

## SUMMARY

Over 20,000 tubers of 6 varieties of potatoes obtained from 10 different states were graded individually for specific gravity. Varietal differences were noted in the specific gravity values. Katahdin showed the greatest

variability and Irish Cobbler and Chippewa the least. The variability in specific gravity as shown by the standard deviations indicates that some lots and varieties are more adaptable than others to commercial separation into different specific gravity lots. The moisture content of the air in the storage rooms is an important factor controlling changes in specific gravity during storage. Potatoes stored at relative humidities of approximately 90 per cent showed slight changes in specific gravity during storage but lower humidities caused noticeable changes. Varieties differed materially in shrinkage and in their changes in specific gravity during storage.

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# ENZYMIC HYDROLYSIS OF POTATOES[1]

E. G. Heisler, R. H. Treadway, Madelyn F. Osborne, and Marian L. McClennan

*Eastern Regional Research Laboratory[2], Philadelphia 18, Pa.*

(Accepted for publication August 23, 1951)

## Introduction

During recent years, cull and surplus potatoes have been used in enormous quantities for alcoholic fermentation and in small amounts for other fermentations. Industry has needed more information on the use of different types of amylases in converting the starch of potatoes to sugars prior to fermentation. Knowledge pertaining to the hydrolysis of potatoes is also necessary to further their use in applications other than fermentations.

This study on enzymic hydrolysis of potatoes was instituted to obtain data on better methods of producing potato hydrolyzates for fermentations and for such uses as adhesives, binders, sizes, thickeners, and glues. Commercial amylase preparations are herein compared in terms of their ability to convert starch into dextrins and sugars. An amylase that gives predominantly dextrins and relatively little sugars would be of value when

[1] Report of a study made under the Research and Marketing Act of 1946.

[2] One of the laboratories of the Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, United States Department of Agriculture.

the starch of potatoes is to be converted into a viscous paste of high cohesive and adhesive strengths for use as an adhesive, for example. On the other hand, an amylase that converts starch primarily to sugars is required when potatoes are used as raw material for fermentations or for preparation of sirups.

## Procedures

*Cooking.* — Washed potatoes were ground in a hammer mill, mixed with 0.1 per cent RHozyme DX[3] liquefying enzyme (based on the weight of starch), and autoclaved. Since the potatoes contained 13 per cent starch on the average, the amount of liquefying enzyme added was ordinarily about 0.013 gram per 100 grams fresh potatoes. Five thousand grams of ground potatoes were used per batch in a 2-gallon glass-lined autoclave equipped with a stirrer. About 1 hour was required to bring the potatoes to 130° C. During this period, the temperature was maintained at 40° to 60° for at least 15 minutes to give the liquefying enzyme a chance to thin the starch. After this, the enzyme was destroyed by raising the temperature to 130°-135°, and holding it for ½ hour. The sole purpose of using the liquefying enzyme was to render the cooked material fluid enough to handle. It was determined that use of this low concentration of enzyme for 15 minutes produced no measurable amount of sugar.

*Hydrolysis.* — After the autoclaving, the cooked potatoes were blown out of the pressure vessel. Portions of 1000 grams were placed in containers held in a constant temperature bath and provided with efficient agitation. The naturally occurring pH of the potatoes (about 5.5) was not altered except when pH was studied as a variable. As soon as the temperature of the cooked potatoes equalled that of the bath, the enzyme (usually 1 per cent by weight based on the amount of starch or what amounted to about 1.3 grams per 1000-gram portion of potatoes) was added in a water solution or suspension. Although this amount of enzyme is greater than that ordinarily recommended by manufacturers, it was not an excessive concentration. Addition of the enzyme was taken as zero time in the hydrolysis. Samples were removed after ½, 1½, 2½ and 4 hours. The 100-ml. samples were placed in a 500-ml. Erlenmeyer flask immersed in a 90° C. bath; enzymic activity was stopped in a few minutes at this temperature. The samples were then ready for analysis.

*Preparation of Sirups.* In the preparation of sirups, hydrolysis was similar to the rest except that higher enzyme concentration was used to assure high conversion to sugars in 1 hour at 55° C. For example, 15 per cent barley malt (based on the weight of starch) was used in one case. The hydrolyzate was filtered and given one activated carbon treatment, after which it was passed through a cation exchange resin column (Amberlite IR-120)

[3] Products mentioned in this paper are not recommended or endorsed by the U. S. Department of Agriculture over other similar products not mentioned.

followed by passage through an anion exchange resin (Amberlite IRA-400) to remove the positive and negative ions of the amino acids, other nitrogen compounds, inorganic salts, and other compounds of the potato. Permutit DR, a decolorizing resin, was as efficient as activated carbon and much more convenient to use[3]. The hydrolyzate was then given another carbon treatment and concentrated under reduced pressure at 35° to 45°.

*Analytical Determinations.*

*Total Solids.* The 15 to 20 gram sample was dried for 2 hours at 65° C. in a mechanical convection oven, followed by drying to constant weight in a vacuum oven at 70°.

*Starch.* The method of Steiner and Guthrie (7) and the shorter method of Clendenning (1), which gave essentially the same result on identical samples, were used.

*Dextrose and Maltose.* The hydrolyzate was prepared for the sugar determinations by use of the A.O.A.C. method (3).

The determinations were made by the Zerban and Sattler modification of the Steinhoff method (8), in which dextrose plus maltose, are determined jointly, dextrose is determined separately, and maltose is calculated by difference.

*Total Sugars.* After the crude hydrolyzate was treated by the previously referred to A.O.A.C. method to obtain the extract, the solution was hydrolyzed with one-tenth of its volume of 10 per cent hydrochloric acid (specific gravity 1.125) and sugars determined as dextrose. (4)

*Total Soluble Carbohydrates.* The sample (15 g.) was extracted by shaking with 500 ml. distilled water at 25° - 30° C. and letting the mixture stand for 1 hour. An aliquot part of the filtered extract was then hydrolyzed as in the total sugars determination. Sugars were determined as dextrose.

*Higher Sugars.* Oligosaccharides other than maltose were calculated by subtracting dextrose plus maltose from total sugars. It is assumed that the reducing power of these higher sugars can be neglected for all practical purposes.

## Data and Discussion

*Amylases Used.* Amylases, like other enzymes, are proteins. They exist in crystalline form when pure, and have high molecular weights. Each amylase has its optimum temperature, pH, and environment. Commercial amylase preparations high in alpha-amylase liquefy starch, producing dextrins quickly. Alpha-amylase is a versatile amylase, however, that not only liquefies starch in its primary phase but also produces sugars by its later action. Beta-amylase produces maltose primarily and does not liquefy starch rapidly.

Twelve commercial preparations, sprouted barley (barley malt), and a

recently developed fungal amylase were evaluated. There was considerable variation in the potency of the preparations. This was expected, since some commercial amylases are extracts of the soluble constituents, whereas others, like sprouted barley, contain a preponderance of non-active material. Some commercial amylases also contain inert diluents.

Table 1 gives the origin and potency of the various preparations in terms of alpha- and beta-amylase units. Alpha-amylase values were obtained by the method of Olson, Evans, and Dickson. (5) The only change made in their procedure was the determination of activity at 30° C., as previously practiced by Sandstedt, Kneen, and Blish (6), instead of at 20°.

This method was originally developed for use with barley malt. Although it has been employed to assay amylases of other origins, the method has limitations with amylases other than malt.

Beta-amylase activity was determined by the method of Kneen and Sandstedt. (2)

TABLE 1.—*Potency of Commercial Amylases*

| Enzyme | Source | Alpha-Amylase Units* | Beta-Amylase Units† |
|---|---|---|---|
| *Group 1, saccharifying:* | | | |
| Sprouted barley | Plant | 25 | 11.4 |
| Malt sirup (Malt Diastase Co.) | " | 100 | Not determined |
| Pancreatin (Takamine Lab., Inc.) | Animal | 375 | ‡ |
| Malt diastase " " " | Plant | 400 | 19 |
| Enzyme MB (Wallerstein Labs.) | Fungus | 2,400 | 0 |
| Clarase (Takamine Lab., Inc.) | " | 4,957 | 0 |
| RHozyme S (Rohm & Haas Co.) | Microorganism | 6,810 | 0 |
| Dextrinase (Takamine Lab., Inc.) | Fungus | 7,525 | 0 |
| Enzyme BS " " " | § | 9,240 | 67 |
| Fungal diastase (Paul-Lewis Labs.) | Fungus | 15,967 | 0 |
| *Group 2, liquefying:* | | | |
| RHozyme DX (Rohm & Haas Co.) | Fungus | 194 | 1.2 |
| Rapidase W3F (Wallerstein Labs.) | Bacterial | 333 | 6.8 |
| Vanzyme 31 (R. T. Vanderbilt Co.) | | 1,695 | 13.3 |

* Grams soluble starch which in the presence of an excess of beta-amylase are dextrinized by 1 gram enzyme in 1 hour at 30° C.
† Grams soluble starch converted to maltose by the beta-amylase of 1 gram enzyme in 1 hour at 30° C.
‡ Result considered doubtful.
§ Source not disclosed by manufacturer.

As exemplified by the typical cases shown in figures 1 and 2, the amylases used in this study fell into two groups: (1) Saccharifying enzymes that give a high sugar concentration, principally maltose as a rule, early in the hydrolysis, and (2) liquefying enzymes that quickly convert starch to dextrins but require considerable conversion time for production of appreciable sugars. For this reason, the amylases are separated into two groups both in table 1 and in the discussion of the hydrolyses.

The results obtained in the enzymic hydrolysis of potatoes did not correlate unusually well with the alpha- and beta-amylase potencies as determined by standard procedures. Table 1 shows that, as expected because of the nature of their hydrolyses, two of the three amylases of Group 2 had low beta-amylase potencies and reasonably high alpha-amylase potencies. Enzymic preparations high in alpha-amylase, however, produced

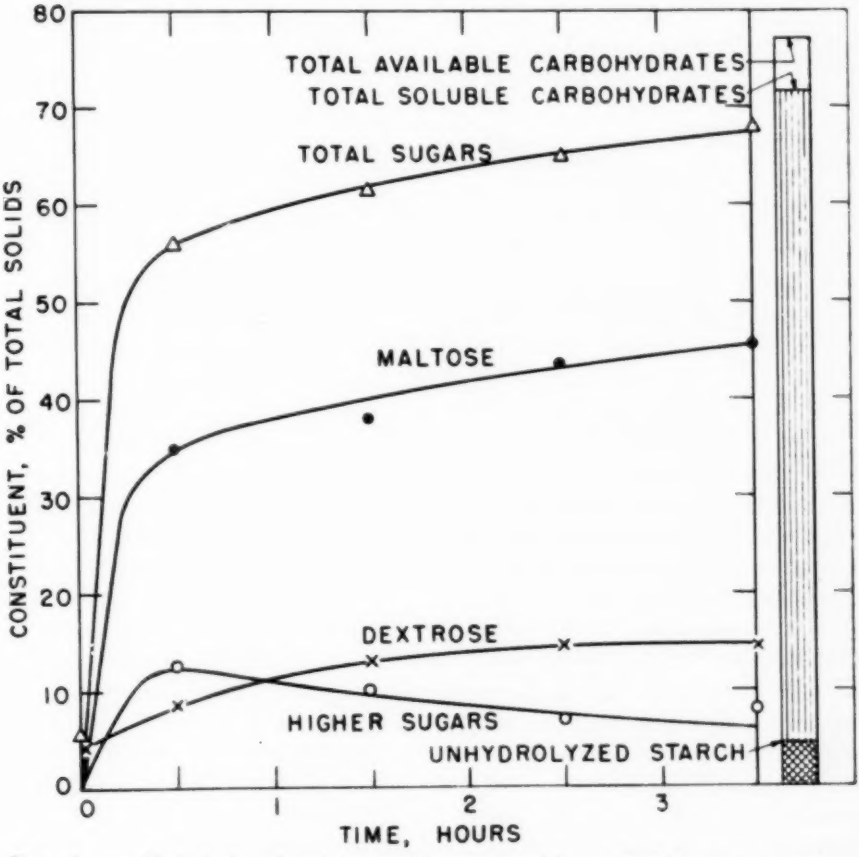


Fig.. 1. — Hydrolysis of potatoes with commercial saccharifying-type amylase (RHozyme S)

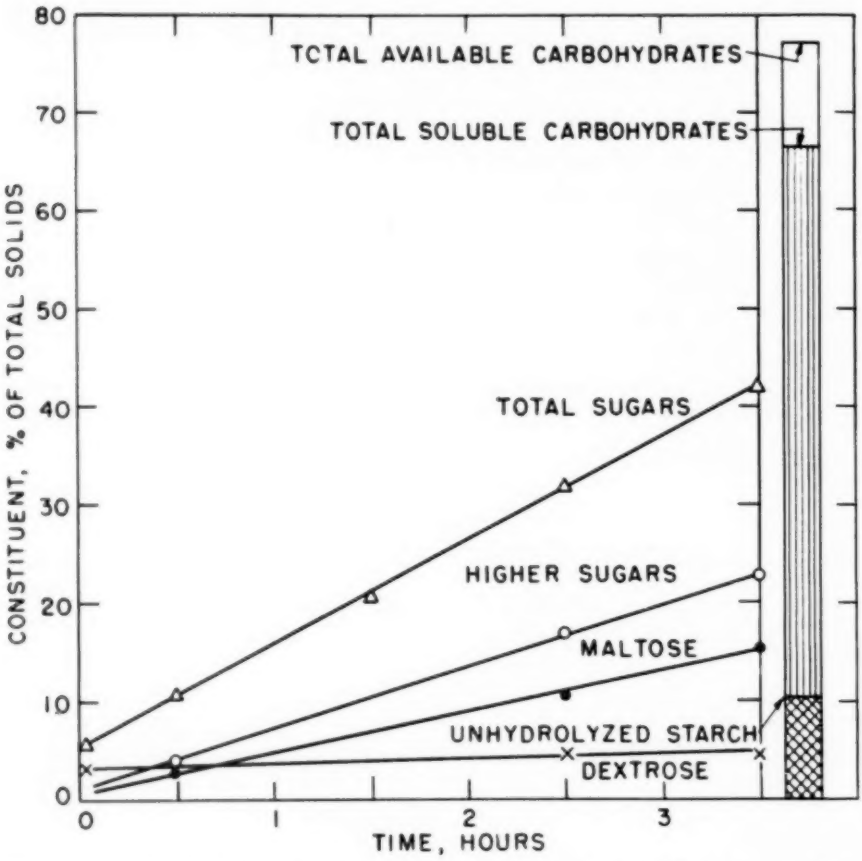


Fig. 2. — Hydrolysis of potatoes with commercial liquefying-type amylase (RHozyme DX)

rapid saccharification, even when beta-amylase was low or *nil*. This bears out the fact that alpha-amylase is a fundamental amylase and, when present in sufficient potency, will bring about saccharification as well as liquefaction.

Dextrinogenic activity is a function principally of alpha-amylase, but alpha-and beta-amylase overlap in their saccharogenic activity. Some of the commercial amylases used undoubtedly contained maltase and other carbohydrases in addition to alpha- and beta-amylase, but it was not an object of this study to characterize them in detail. No attempt is made, therefore, to explain why Group 1 and Group 2 commercial amylases gave different types of hydrolysis.

*Hydrolyses by Group 1 Amylases.* All the amylases of Group 1 rapidly produced sugars, of which maltose predominated. Since similar results

TABLE 2.—*Hydrolysis of potatoes by various commercial saccharifying amylases. Carbohydrate composition of hydrolyzate after 30 minutes hydrolysis 1 per cent enzyme; 55° C.*

| Enzyme | Dextrins | Total Sugars | Higher Sugars | Maltose | Dextrose |
|---|---|---|---|---|---|
| Sprouted barley* | 13 | 54 | 6 | 43 | 5 |
| Malt sirup | 28.5 | 41.5 | 2.5 | 33.5 | 5.5 |
| Pancreatin | 17.5 | 55 | 7 | 39.5 | 8.5 |
| Enzyme MB | 23.5 | 45 | 16 | 22.5 | 6.5 |
| Clarase | 13.5 | 52.5 | 19.5 | 27.5 | 5.5 |
| RHozyme S | 15.5 | 56 | 12.5 | 35 | 8.5 |
| Dextrinase | 14.5 | 61 | 11 | 39 | 11 |
| Enzyme BS | 21. | 50.5 | 24 | 20 | 6.5 |
| Fungal diastase | 9 | 65.5 | 13.5 | 45.5 | 6.5 |

* 15 per cent by weight, based on starch weight.

were obtained for all 10 amylases of this type, one of the group (RHozyme S) was selected as typical and its curves illustrating rate of sugar formation are presented. Figure 1 shows the carbohydrate composition of the hydrolyzate during 3½ hours of hydrolysis. Table 2 gives compositions of the hydrolyzates obtained with the various saccharifying amylases.

These enzymes produced total sugars at a high rate initially and then at a slower rate after one-half hour or so. No attempt was made to investigate the kinetics of the hydrolyses, since the enzyme mixtures in commercial amylase preparations would make this too complicated. The "higher sugars" (unknown oligosaccharides) produced in the hydrolysis, although present in smaller amount than maltose, were generally found in greater concentration than dextrose at the ½-hour sampling time. They usually decreased during the following hour, as dextrose increased. The values of the higher sugars typically assumed a low level after 3 or 4 hours' hydrolysis, at which time the sugars were nearly all maltose and dextrose.

The total soluble carbohydrates values were always a little less than the total available carbohydrates (starch plus total sugars) values. A small amount of unhydrolyzed starch was invariably left at the end of a conversion and remained unextracted during the determination of total soluble carbohydrates. The amount of unhydrolyzed starch found by direct determination was frequently equal to the difference between total available and total soluble carbohydrates, within experimental error.

*Hydrolyses by Group 2 Amylases.* RHozyme DX was the slowest sugar

TABLE 3.—*Hydrolysis of potatoes by commercial liquefying amylases. Carbohydrate composition of hydrolyzate after 30 minutes hydrolysis 1 per cent enzyme; 55° C.*

| Enzyme | Dextrins | Total Sugars | Higher Sugars | Maltose | Dextrose |
|---|---|---|---|---|---|
| RHozyme DX | 56 | 10.5 | 4 | 3.5 | 3 |
| Rapidase W3F | 40 | 23.5 | 13.5 | 5.5 | 4.5 |
| Vanzyme 31* | 26 | 48.5 | 24.5 | 19 | 5 |

* This enzyme is sold by the manufacturer as a starch liquefying agent for production of adhesives and sizes. Conditions used in our hydrolysis, which resulted in high sugar formation, were considerably different from the enzyme concentration, pH, and temperature recommended by the manufacturer.

producer of all the amylases studied. Figure 2 gives the rates of formation of sugars in the hydrolysis of potatoes by this enzyme. The per cent sugar-time relationships are nearly linear. Table 3 shows the composition of the hydrolyzates obtained with three liquefying amylases.

Rapidase W3F also produced sugar slowly, although it had high dextrinizing power. Vanzyme 31 was used at 1 per cent concentration based on the starch, at pH 5.5, and at 55° C. in order to compare its activity with other commercial amylases under the same conditions. It produced much sugar, however, likely because the recommended conditions for using it as a liquefying amylase are far different from those we employed. The manufacturer's recommendations call for a lower enzyme concentration, a higher pH, and a considerably higher temperature. The amount of higher sugars formed in the hydrolysis by each of these three amylases was roughly equal to the sum of maltose and dextrose. The liquefying enzymes differed in action from Group 1 amylases in that the concentration of higher sugars increased continuously during the 4-hour conversion. Dextrose values remained almost constant at low levels.

*Action of a Commercial Hemicellulase.* A sample of hemicellulase obtained from the Paul-Lewis Laboratories saccharified potatoes well. In one experiment, the potatoes originally contained 67.5 per cent starch and 4.8 per cent total sugars on the dry basis. After 4 hours' hydrolysis at 55° with 1 per cent added hemicellulase (based on weight of starch), the hydrolyzate consisted of the following (per cent, dry basis): Total soluble carbohydrates (expressed as dextrose equivalent after acid hydrolysis of solubles) 66.5; dextrose 9.4; maltose 19.9; higher sugars 13.4; dextrins 21.4.

A mixture of 1 per cent hemicellulase and 1 per cent amylase (starch weight basis) added to cooked, ground potatoes gave a significantly higher soluble carbohydrates value than that accounted for by adding the starch and total sugars values. Clearly the hemicellulase attacked higher carbo-

hydrates that are untouched by amylases. The hydrolyzate was not clear, however. Even if all the hemicellulose were converted to soluble form, the corky skin tissue (and perhaps some cellulose) would remain undissolved.

*Effect of the Non-Amylaceous Constituents of the Potato on Enzymic Hydrolysis of Starch.*

To determine how hydrolysis of the starch in potato differs from hydrolysis of pure potato starch (in distilled water), several amylases were used in comparative conversions of both substrates. Since amylases are commonly affected by the presence of various substances in the medium, it was expected that the nitrogen compounds, salts, and acids of the potato would alter the results. The pH was about 5.5, and the starch concentration was the same in both the aqueous starch and potato media. Data for some of the Group 1 and Group 2 amylases are given in table 4. To conserve space, data are given for 4 hours' conversion only and for only two enzymes in each classification. The amount of starch solubilized was greater when pure starch was used. Group 1 amylases converted a higher proportion of pure starch than starch in potato to dextrose. The saccharifying amylases gave about the same conversion to maltose for the two substrates. Group 2

TABLE 4.—*Comparative hydrolyses of potatoes and pure starch by commercial amylases 1 per cent amylase; 55° C.*

| Enzyme | Substrate | Per cent Starch Originally Present* | Per cent Starch Converted after 4 Hours to — Total Soluble Carbohydrates | Maltose | Dextrose | Higher Sugars |
|---|---|---|---|---|---|---|
| *Group 1* Saccharifying Amylases | | | | | | |
| Enzyme MB | Potato | 64.1 | 85.2 | 59.7 | 8.8 | 6.0 |
| | Starch | 98.5 | 94.1 | 53.4 | 23.3 | —†† |
| RHozyme S | Potato | 64.0 | 92.8 | 65.1 | 15.6 | 11.2 |
| | Starch | 98.5 | 100.9 | 61.7 | 19.8 | —— |
| *Group 2* Liquefying Amylases | | | | | | |
| RHozyme DX | Potato | 69.2 | 79.6 | 22.8 | 1.6 | 33.8 |
| | Starch | 98.5 | 90.4 | 33.9 | 2.6 | —— |
| Rapidase W3F | Potato | 61.7 | 81.7 | 26.3 | 0 | 32.2 |
| | Starch | 98.5 | 91.0 | 31.8 | 0 | —— |

* Expressed on dry basis.
† Not determined.

amylases differed from Group 1 in that they produced little or no dextrose when acting on either substrate; they converted a higher amount of pure starch than starch in potato to maltose.

*Effects of pH, Temperature, and Enzyme Concentration*

We have found that little or nothing is gained in enzyme activity by adjusting a potato slurry from its natural pH (about 5.5) to the pH recommended by the amylase manufacturer for starch hydrolysis. Not only rate of hydrolysis but also composition of the hydrolyzate is nearly the same, whether or not the conversion takes place at the pH of the potato or at the recommended pH; sometimes addition of sodium hydroxide or hydrochloric acid has a detrimental effect. Owing to the buffering action of the salts and nitrogen compounds of potato juice, a relatively large amount of either acid or alkali is required for changing the natural pH. The optimum pH of the various amylases used, as recommended by the manufacturers, ranged from 4 to 7.5.

Most commercial amylases, except those of bacterial and pancreatic origins, have optimum temperatures near 55° C., the temperature at which comparisons were made. Only minor changes, therefore, have usually resulted when hydrolysis was carried out at the recommended temperature for a preparation instead of at 55°. A commercial fungal diastase (product of the Paul-Lewis Laboratories), having a recommended optimum temperature of 45°, gave virtually the same results at 55°. Even a wide variation in temperature usually made little difference in the hydrolysis. Another fungal diastase (Dextrinase), with optimum temperature 40°, gave similar results at 65°.

The composition of a potato hydrolyzate varied within wide limits, however, with changes in enzyme concentration. Higher enzyme concentration does not simply make the hydrolysis proceed faster in a pattern similar to that obtained at low amylase concentration. For example, dextrinase gave a family of smooth curves for rate of dextrose formation with increasing amounts of enzyme (0.5 to 2.5 per cent). The maltose concentration increased continuously with time when 0.5 per cent of this amylase was present, but passed through maxima with 1.5 and 2.5 per cent enzyme. Maltose concentration, in the latter case, was reduced during the later stages of the hydrolysis, as it was split to dextrose.

## Sirups

Glucose sirup has been prepared commercially from potato starch by acid hydrolysis in a manner similar to production of corn sirup. One or two companies that have made sirup by enzymic hydrolysis of starch in the presence of other constituents of the potato, however, have met with difficulties in obtaining a clear, light-colored product. As a part of our

study on enzymic hydrolysis of potatoes, sirups were produced directly from potatoes in the hope that either an improved process or a product of unique flavor could be developed.

Using the general methods described in this paper, we employed a commercial fungal amylase (Paul-Lewis Laboratories) to convert ground, cooked potatoes to a hydrolyzate of high sugar content. A sweet, amber-colored sirup of pleasing taste was produced.

Bland, virtually colorless sirups of high clarity and sweetness were produced by either barley malt or commercial amylases. The potato hydrolyzates were concentrated after they had been treated with activated carbon and ion exchange resins. Analyses showed that the nitrogen contents of these sirups were low. It is likely that the organic acids and inorganic salts contents were also low after the ion exchange treatments. As far as we are aware, this is the first preparation of a white, bland sirup by enzymic hydrolysis of potatoes. The flavor of potato sirup is similar to that of ordinary corn sirup.

Several attempts were made to prepare an acceptable potato sirup by using amylase from *Aspergillus niger* NRRL 330[4], cultured *in situ*[5], for the starch conversion. The sirups were of poor flavor, perhaps because of acid produced by this strain of *A. niger*. Probably other types of fungal amylase would be more suitable for producing sirups in this way.

## Summary and Conclusions

A number of commercial amylases have given rapid conversion of starch in potato to sugars. Most of the amylases produced nearly their maximum amount of sugar in ½ hour at 55° C. with 1 per cent enzyme, based on weight of the starch.

Maltose was the principal sugar produced by all amylases. Unknown oligosaccharides were produced in the hydrolyses, generally in greater amount than dextrose in the early phase. These oligosaccharides, which may well be unfermentable in several of the most common types of fermentations, can be reduced to an insignificant amount either by carrying out the conversion for 3 or 4 hours or by using a higher concentration of amylase.

There was not much correlation between the alpha- and beta-amylase potencies of the commercial amylases and their performance in hydrolyzing potatoes under the conditions used. As expected, the slow sugar-forming amylases were relatively low in beta-amylase potency, but preparations

[4] This strain of *A. niger* was found by the Northern Regional Research Laboratory, Peoria, Ill., to be a good producer of amylases.

[5] The culture of *A. niger* NRRL 330 on potatoes and the conversion of starch to sugars were carried out simultaneously by E. A. Weaver of the Eastern Regional Research Laboratory.

having little or no beta-amylase but powerful alpha-amylase activity produced sugars rapidly.

The amylases used converted more pure starch than starch in potato to sugars, under the same conditions.

It apparently is unwise to change the naturally occurring pH of potatoes to be enzymically hydrolyzed. Results obtained with pH adjusted (by sodium hydroxide or hydrochloric acid) to the optimum recommended by amylase manufacturers were no better, and sometimes worse, than the corresponding control.

Recommended temperatures for pure starch conversions by commercial amylases did not apply in a critical manner to the potato hydrolyses.

The composition of potato hydrolyzates varied widely with changes in the enzyme concentration.

Virtually colorless sirups of high degree of sweetness and pleasant taste have been prepared from purified potato hydrolyzates.

## Acknowledgments

The authors wish to express their appreciation to the various companies who supplied generous samples of the amylases used in this study. We are also indebted to C. H. Fisher and E. Yanovsky for their advice and encouragement.

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# BLIGHT-RESISTANT POTATO VARIETIES CAN SAVE COPPER AND CUT COST OF PRODUCTION

ROBERT V. AKELEY,[1] P. M. LOMBARD,[2] AND F. J. STEVENSON[3]

*Bureau of Plant Industry, Soils, and Agricultural Engineering, United States Department of Agriculture, Beltsville, Md.*

(Accepted for publication September 11, 1951)

In these days of national emergency more attention should be given to the growing of crops that are resistant to disease. Some of the potato varieties that have been released in recent years, such as Kennebec, Pungo, Cherokee, Essex, Ashworth, and others are highly resistant to, if not immune from the common races of the late blight fungus, *Phytophthora infestans* (Mont.) DeBy. At present these varieties can be grown without the application of fungicides or with fewer applications than are given to any of the old standard varieties. If all the potato acreage in the United States were planted to varieties that are highly resistant to late blight much of the 50,000,000 pounds of copper sulfate ,which, according to estimates, is used to protect the crops from blight, could be saved; and copper is on the critical list of the defense program.

In Maine, Kennebec yields as well without the application of a fungicide as it does when sprayed with bordeaux at weekly intervals throughout the growing season. This is evident from tests made on the Aroostook Farm, Presque Isle, Maine, in 1949 and 1950. In 1949 a test was made to study the effect of various sprays on Kennebec, Menominee, Green Mountain, and Katahdin. Four treatments were used: bordeaux, bordeaux + DDT, DDT, and no spray. The design of the experiment was a split block 4 x 4 latin square. The four varieties were planted in each block with four border rows of a blight susceptible variety, Green Mountain, on each side to serve as a medium on which the blight fungus would grow rapidly and to protect the varieties under test from spray drift. Each plot consisted of 25 hills planted 12 inches apart, and the rows were planted 34 inches apart. The plots were inoculated with the late blight fungus on July 14, and a heavy infestation of the disease was soon prevalent on the susceptible varieties. The first sprays were applied on June 29 and repeated at weekly intervals throughout the remainder of the growing season.

The data for the Kennebec variety for 1949 (Table 1) indicated that bordeaux decreased rather than increased the yields. The mean yield for the plots sprayed with bordeaux was 11 bushels per acre less than that for the

[1] Horticulturist.
[2] Collaborator.
[3] Principal geneticist.

TABLE 1.—*Effect of sprays on the yields of four varieties of potatoes grown on Aroostook Farm, Presque Isle, Maine, in 1949.*

| Spray | Yield per Acre of U. S. No. 1 Potatoes | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Kennebec | | Menominee | | Green Mountain | | Katahdin | |
| | Bus. | Per cent | Bus. | Per cent | Bus. | Per cent | Bus. | Per cent |
| Bordeaux | 464 | 96 | 379 | 94 | 389 | 95 | 360 | 96 |
| Bordeaux + DDT | 507 | 96 | 416 | 96 | 472 | 96 | 449 | 97 |
| DDT | 599 | 96 | 355 | 95 | 315 | 93 | 357 | 95 |
| None (check) | 475 | 96 | 347 | 93 | 274 | 92 | 292 | 93 |

L.S.D. 5-per cent level for two varieties given the same treatment, 66.0 bus.
L.S.D. " " " for treatment means for same or different varieties 67.8 bus.

unsprayed check plots. However, there was a highly significant difference —124 bushels per acre— between the plots sprayed with DDT and the checks. The plots of Kennebec sprayed with DDT alone yielded 599 bushels per acre but when sprayed with bordeaux + DDT the yield was 507 bushels per acre. This is a highly significant difference and might indicate that the bordeaux had a depressing effect on the yield of this variety. The high yields produced by the plots sprayed with DDT could not indicate a fungicidal effect, as has been supposed by some, since none of the Kennebec plots was infected with blight. Furthermore, the difference could not be accounted for by assuming that the DDT kept the plants growing later in the season since all of them were planted on the same date and were growing vigorously when killed by frost on September 10. The control of insects by DDT was no doubt responsible for the results.

The Menominee is somewhat resistant to blight, being intermediate between Kennebec and Green Mountain in its reaction. In the case of Menominee there were no significant differences in yield between the check plots, the plots sprayed with bordeaux, and the plots sprayed with DDT. There was a slight and significant difference between the yield of the check plots and that of the plots sprayed with bordeaux + DDT.

The blight-susceptible varieties, Green Mountain and Katahdin, gave the highest yields when sprayed with bordeaux + DDT. Green Mountain sprayed with bordeaux alone gave a significantly higher yield than when it was sprayed with DDT alone. On the other hand, there was no significant difference between the yields of the Katahdin plots that received these two spray treatments.

The test was repeated in 1950 with a number of changes. Sebago was used instead of Menominee, the number of plants per plot was increased from 25 to 30, and the plants were spaced 10 instead of 12 inches apart.

The same treatments — bordeaux, bordeaux + DDT, DDT, and no spray — were used. The design of the experiment, a split block 4 x 4 Latin square, was the same. The four varieties, Kennebec, Green Mountain, Sebago, and Katahdin, were planted in each block with four border rows of Teton on each side to protect the varieties under test from spray drift and to provide a medium on which late blight would develop rapidly. All plots were inoculated with the late blight fungus on July 13, and soon there was a heavy epidemic on the border rows of Teton which were not sprayed, and on the plots of the susceptible varieties that were not sprayed with bordeaux.

The yield data for this test are given in table 2.

Again, as in 1949, there was no significant difference between the yields produced by the Kennebec plots that received no spray and those that were sprayed with bordeaux alone. The highest yields produced by this variety were from plots sprayed with DDT alone. There was no significant difference between the yields of the plots sprayed with bordeaux + DDT and of those sprayed with DDT alone, indicating that in 1950 bordeaux had no effect when used on Kennebec. The plots of Green Mountain, Sebago, and Katahdin sprayed with bordeaux + DDT produced the highest yields, with bordeaux alone ranking second. The low percentages of U. S. No. 1 tubers produced on the DDT and check plots of Green Mountain were due to some extent to tuber rots. No rots were found in Kennebec and Sebago.

The tests for 2 years indicate that when late blight is prevalent in epidemic proportions in Maine, Kennebec will yield satisfactorily without fungicide spray; DDT sprays to control insects will increase yields but

TABLE 2.—*Effect of sprays on the yields of four varieties of potatoes grown on Aroostook Farm, Presque Isle, Maine, in 1950.*

| Spray | Yield per Acre of U. S. No. 1 Potatoes | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Kennebec | | Green Mountain | | Sebago | | Katahdin | |
| | Bus. | Per cent | Bus. | Per cent | Bus. | Per cent | Bus. | Per cent |
| Bordeaux | 569 | 98 | 522 | 94 | 455 | 92 | 509 | 93 |
| Bordeaux + DDT | 658 | 98 | 571 | 93 | 471 | 93 | 555 | 94 |
| DDT | 665 | 97 | 359 | 78 | 403 | 91 | 441 | 86 |
| None (check) | 540 | 97 | 332 | 79 | 384 | 89 | 403 | 86 |
| Mean | 608 | 98 | 446 | 86 | 428 | 91 | 477 | 90 |

L.S.D. 5 per cent level, for two varieties given the same treatment, 69.5 bus.
L.S.D. " " " for two treatment means for the same or different varieties 65.9 bus.

bordeaux seems to be of no value. Susceptible varieties should be sprayed with fungicides and insecticides.

## Discussion

Kennebec has been a failure in a number of tests in the South although it maintained its resistance to late blight. In some sections, especially where early blight is prevalent, it would pay to spray this variety with fungicides. On the other hand, Kennebec has produced high yields of excellent market and cooking qualities in Maine even when inoculated with late blight spores. As grown on the plots on Aroostook Farm, the tubers are excellent for baking, boiling, frying, French frying, and chipping. Kennebec is as near an all-purpose potato as any now grown commercially.

If varieties with the blight resistance of Kennebec were more generally grown there would be a saving not only in critical copper compounds but also in the cost of producing the potato crops. One grower estimated that if he grew 40 acres of Kennebec without spraying with fungicides but using DDT alone he could save about $2,500. In addition, his yield and crop value would be greater than if he grew the same acreage of Green Mountain or Irish Cobbler.

Some alarm has been expressed over the fact that Kennebec is not immune from the more virulent races of the late blight fungus and that as soon as these become prevalent in Maine this variety will lose its resistance. This may be true but it may also be a long time before the virulent races are distributed through the potato-growing areas. Salaman (1) showed that one resistant seedling grown in the kitchen garden at Barley, England, for 17 consecutive seasons never showed the least sign of infection with blight. In the same book the following statement was also made: "By 1926 I was in possession of over a score of seedling varieties endowed with reasonably good economic characters which, no matter what their maturity appeared to be immune to blight. A few of these were sent to Ireland where they have been growing for over twenty years in the open and have retained a high degree of resistance to attack throughout."

If the varieties in the United States maintained their resistance for 20 years the country will then be repaid for the money it has invested in potato breeding. At this time, varieties will be available that are immune from the attacks of the present more virulent races. A number of selections have already been produced and are now being tested for yield, adaptation, and market and cooking qualities. Some of these are most promising from the commercial standpoint.

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# WASHINGTON CERTIFIED SEED POTATOES

LOUIS W. KING[1]
*State Potato Specialist, Department of Agriculture, Bellingham, Wash.*

(Accepted for publication, July 27, 1951)

The Certified Seed Potatoes of Washington have been tested either in winter plots in California or in the greenhouses in Mount Vernon, Washington. Samples have been taken from *all* the certified fields to arrive at the percentage of disease in these stocks. In 1945 we were using calcium arsenate for the control of the potato flea beetle. The aphis infestation in the plots during 1945 was very heavy, and as can be seen from the enclosed table, many of the fields had a high incidence of disease, despite careful dusting and roguing. The season of 1945 was a disastrous year for the Washington Certified Seed Potatoes and much of the seed did not give a satisfactory clean crop. More than 79 per cent of the White Rose variety showed more than 3 per cent disease. The Netted Gem seed too, that had been largely purchased from Canada, did not give as clean a certified seed as the trade required.

The following year, after many of the growers had used DDT, there was quite an improvement. In the 1947 crop after two years, 86 per cent of the White Rose variety showed less than 3 per cent diseased. Since that time each succeeding crop has gradually improved until the 1950 crop showed 100 per cent of the White Rose variety with 3 per cent disease; and 96 per cent of the Netted Gem with less than 2 per cent. Fifty-seven per cent of all the White Rose crops and 78 per cent of the Netted Gem showed no disease whatever. The State of Washington can now be proud of a job well done by restoring the industry to the point where the seed is very dependable. This cuts the roguing cost of the fields down to a minimum. In fact, it is getting to be monotonous work to rogue a field of seed in this area.

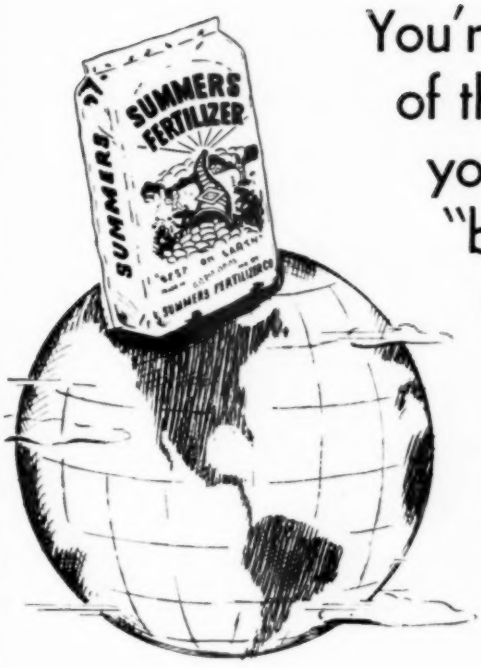
Whatcom County produces about 90 per cent of the Certified Seed in the State of Washington, 90 per cent of which show less than 1 per cent disease and 100 per cent less than 3 per cent, with no leaf roll in any of the acreages where the White Rose variety was grown. We have a very high class foundation seed potato. This increase may be attributed to the fact that we have controlled the insects in 100 per cent of the seed and commercial fields. In this way we have been able to control the spread of disease by controlling the vector. This would not be possible were it not for the fact that all the fields have insect control. The only exception to this is prevalent in a few certified fields in Eastern Washington and it is in those fields, where we found more than 3 per cent disease.

[1] State Potato Specialist.

In table 1 data are given on two varieties only, White Rose and Netted Gem. We also grew Burbank, Pontiac, Gold Coin, Sebago, Bliss Triumph, Irish Cobbler, Beauty of Hebron, Kennebec, Early Rose and Katahdin, in smaller acreages, mostly for home garden planting. The data on these have not yet been compiled, but they are fairly comparable to the percentages of the two varieties shown.

TABLE 1.—*Insect transmitted disease in Washington seed potatoes based on winter test plot readings*

| Classification of Disease Intensities | Percentages with Different Intensities of Disease | | | | | |
|---|---|---|---|---|---|---|
| | 1945 Crop | 1946 Crop | 1947 Crop | 1948 Crop | 1949 Crop | 1950 Crop |
| | | | | White Rose | | |
| No disease observed | 9.37 | 21.90 | 23.33 | 50.00 | 37.80 | 57.40 |
| Less than 1 per cent | 10.94 | 45.10 | 41.67 | 72.22 | 65.85 | 90.74 |
| Less than 2 per cent | 10.94 | 59.70 | 65.00 | 83.33 | 85.36 | 98.11 |
| Less than 3 per cent | 12.50 | 68.20 | 86.67 | 89.32 | 92.68 | 100.00 |
| More than 3 per cent | 79.69 | 31.80 | 11.67 | 10.18 | 7.32 | 0 |
| More than 5 per cent | 65.62 | 20.73 | 1.66 | 3.70 | 6.09 | 0 |
| | | | | Netted Gem | | |
| No disease observed | 35.71 | 50.00 | 21.43 | 32.36 | 45.45 | 77.77 |
| Less than 1 per cent | 42.85 | 53.12 | 50.00 | 50.00 | 72.72 | 88.88 |
| Less than 2 per cent | 57.14 | 65.12 | 57.14 | 76.47 | 87.87 | 96.29 |
| Less than 3 per cent | 71.47 | 78.13 | 64.28 | 100.00 | 96.96 | 96.29 |
| More than 3 per cent | 28.53 | 21.87 | 35.72 | —— | 3.03 | 3.70 |
| More than 5 per cent | 14.28 | 9.37 | 28.51 | —— | 3.03 | 0 |

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